The appearance of polar bodies in the spermogenous tissue of Ricciocarpus natans (L.) Corda

RUTH S. ATWELL

(WITH PLATE 8)

The presence of polar or centrosome-like bodies in spermogenous cells and their possible function and origin in the cell continues to be the subject of much discussion and interest among the cytologists working with Bryophyta. Such bodies have been noted by Ikeno (2) and Mottier (4) in *Marchantia* and regarded by them as true centrosomes. Lewis (3) observes them in *Riccia natans* L. (*Ricciocarpus natans* Corda) and in *Riccia crystallina* L. but does not regard them as true centrosomes since they arise *de novo* with each division. Miss Black (1) observes that the last division in the spermogenous tissue of *Riccia Frostii* Aust., the diagonal division, is accompanied sometimes by granules at the poles.

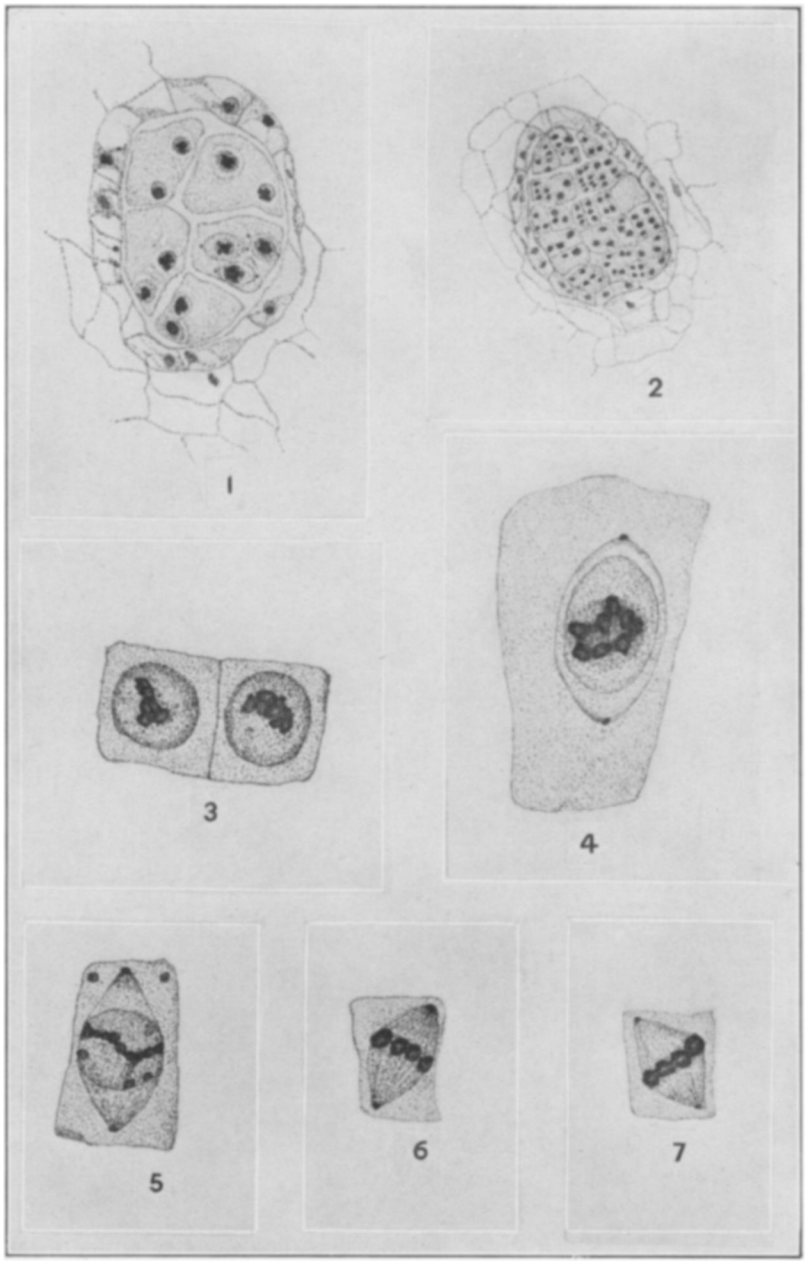
For this preliminary study of spermatogenesis in certain Hepaticae, *Ricciocarpus natans* (L.) Corda furnished the material from which slides were prepared and certain facts noted. These may be of interest by way of comparison or confirmation of the conclusions formed by workers in other related species. Material of *Ricciocarpus natans* was gathered near the Skokie Marsh, Cook County, Illinois, during June and July, 1913, and was fixed in chromic-osmic-acetic solution. The sections were stained with analin safranin and gentian violet. Occasionally orange G was also used. Deep staining followed by washing was necessary as it was difficult otherwise to differentiate the cytoplasmic structures.

As has been observed by others, it was noted that nearly all the stages of karyokinesis could be found in different segments of one of the nearly mature antheridia. All the cells of each segment, as marked off by the first divisions of the antheridium, contained nuclei in the same stage of development. Thus in large antheridia many successive stages of division were found. Fig. 2 shows such an antheridium.

In the earliest prophase of division noted, the chromatin seems to be arranged in a mass of irregular lumps, surrounded by a homogenous material which seems denser next to the nuclear membrane and which has a staining reaction quite similar to the surrounding cytoplasm; no nucleolus is distinguishable; the nuclear membrane is pronounced, the cytoplasm finely and evenly granular. FIG. 3 shows cells typical of this condition. Centrosome-like bodies appear in cells of younger antheridia as well as in older ones. They are also present in cells of both old and young antheridia during the earlier as well as the diagonal division. FIG. 1 shows a young antheridium in one of the segments of which these bodies were observed in connection with the three nuclei of that segment. One of these cells is drawn under higher magnification in FIG. 4. FIG. 6 represents these bodies in a cell of a much older antheridium when the spindle and nucleus are in a more advanced stage than in FIG. 4. The bodies are distinct and cannot be explained as granules appearing accidentally for they appeared quite constantly. They do not seem to be permanent organs as they arise and disappear with each new division.

The origin of these bodies was not taken up. However, their position at the poles and their direct connection with the spindle suggest that they may represent an important factor in the formation of the spindle. Lewis describes these bodies as first appearing at a little distance from the nuclear membrane which elongates as it approaches them. FIGS. 4 and 5 are typical, and show the bodies as entirely outside and free from the nuclear membrane. Kinoplasm is collected about and extending away from them in cap- or cone-like form outlining the spindle, which is apparently produced from the kinoplasm and consists of a few thick fibers converging at the poles. The bodies lie in the position of true centrosomes.

The chromosomes are formed from the central mass of nuclear material and become arranged in the nuclear plate. FIGS. 6 and 7 show them in this position during the diagonal division. They split and pass to the poles. The polar bodies are always present in this last division. No stages of anaphase were observed, probably due to the rapid change of the position of the chromosomes from the plate to the poles. FIG. 7 shows the chro-



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mosomes splitting. Occasionally granules other than those appearing at the poles are observed in the cytoplasm, as shown in FIG. 5.

Mottier and Ikeno, who observed true centrosomes in plants, report that they are formed from a single body dividing in two. In these observations on *Ricciocarpus natans* the bodies seem to appear at once in the polar position with each new division. Whether or not they persist in the spermatid after the diagonal division and may later become blepharoplasts has been discussed by Black (1), Lewis (3), Wilson (5), Woodburn (6), and others. This study was not carried far enough to observe these late stages in *Ricciocarpus natans*.

NORTHWESTERN UNIVERSITY
EVANSTON, ILLINOIS

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Explanation of plate 8

All drawings were made with the aid of a camera lucida. A Spencer microscope was used with apochromatic objectives, 1.5 mm. oil immersion, 3 mm., 16 mm., and compensating oculars 6 × and 12 ×. FIGS. 1 and 2 were drawn at stage level and all others at table level.

FIG. 1. Young antheridium. One segment showing three nuclei with polar bodies. × 1,600.

FIG. 2. Older antheridium. × 150.

FIG. 3. Two cells in early prophase. The chromatin of the nucleus is seen in an irregular mass. × 3,200.

FIG. 4. Cell from segment in FIG. 1, the chromatin in a central mass, the bodies at the poles with kinoplasm extending from them outlining the spindle. × 3,200.

FIG. 5. Cell from an older antheridium. The chromatin is in an early spireme form and the spindle is more fully developed. × 3,200.

FIG. 6. The oblique spindle with chromosomes in equatorial plate, the spindle fibers terminating in the polar bodies. × 3,200.

FIG. 7. Similar to FIG. 6, the chromosomes splitting. × 3,200.